Characterization of DNA Polymorphisms in Three Populations of Hereford Cattle and Their Associations with Growth and Maternal EPD in Line 1 Herefords[1,2]

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ABSTRACT: Three populations of Hereford cattle differing in inbreeding levels and genetic potential for growth were genotyped for seven DNA polymorphisms. The populations were compared to determine differences in allele frequency and genetic variation. Significant differences in allele frequency among the populations were found at six of the seven polymorphisms genotyped, and average genetic variation differed as expected when inbreeding levels were considered. Effects of several polymorphisms on growth and maternal EPD were evaluated for one population (Miles City Line 1 Herefords) using regression analysis. Substitution of a B allele for an A allele of the kappa-casein polymorphism accounted for significant decreases in direct birth weight and maternal 180-d gain from birth to weaning EPD, explaining 15% and 8%, respectively, of EPD variability. Several other significant effects accounting for small portions of EPD variability were also detected.

Key Words: DNA, Polymorphism, Gene Frequency, Hereford, EPD, Markers

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Introduction

Population genetic theories are derived from knowledge of the segregation of genes and their inheritance from parent to offspring. With the exception of single polymorphic genes that produce distinguishable phenotypes (Ibsen, 1933) and protein variants separable by gel electrophoresis (Aschaffenburg and Drewry, 1957; Aschaffenburg, 1961; Wake and Baldwin, 1961), it has been difficult to evaluate these theories for specific loci in bovine populations. However, DNA markers allow genotypes of individuals to be determined at many loci for many species (O'Brien and Graves, 1991), enabling population parameters such as allele and genotype frequencies to be estimated. This information will allow comparisons of frequencies among populations and estimates of genetic variation at the DNA level to be made. These factors may reveal differences in genetic backgrounds of populations that contribute to phenotypic variability.

DNA polymorphisms are also potentially useful as markers of quantitative trait loci (**QTL**) (Soller and Beckman, 1983). Studies have investigated polymorphisms within functional genes to determine whether the polymorphisms could be useful markers of QTL (Lunden et al., 1993; Bovenhuis and Weller, 1994; Rothschild et al., 1994). Candidate genes were selected in these studies because of their biological significance on the quantitative traits of interest.

The objective of this study was to compare population genetic parameters in three unique populations of Hereford cattle differing in average levels of inbreeding and in genetic potential for growth at polymorphisms within seven genes representing a variety of

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locus types. In addition, the effects of five polymorphisms on growth and maternal EPD were evaluated in one population (Miles City Line 1 Herefords) as an example of the candidate gene approach to identify molecular markers of QTL.

Materials and Methods

Populations

The populations involved in this study are unique to the Hereford breed because of differences in their inbreeding levels and genetic potentials for growth. In addition, these populations represent gene pools that have made significant contributions to the Hereford breed.

Lents Anxiety 4th Herefords. The Lents Anxiety 4th Herefords (Indiahoma, OK) have been maintained as a closed population with an emphasis on linebreeding to the bull Imperial Lamplighter, a descendant of Anxiety 4th. The Lents herd was established in 1944 with the bulls Lord Lamplighter 8th and 3 Imperial Lamplighter 49th, who were grandson and great-grandson, respectively, of Imperial Lamplighter (Lents, 1991). The effective population size in the Lents Anxiety 4th herd is approximately 17, as estimated from the equation $4N_mN_f/(N_m + N_f)$ (Falconer, 1989). Significant inbreeding has accumulated in the Lents herd, with an average inbreeding of .54 calculated for herd sires. This herd is unique to most Hereford populations because intentional selection pressure for increased size has not been practiced, and because it represents a gene pool that was influential during the early history of the Hereford breed. Cattle from the Lents Anxiety 4th herd involved in this study included eight herd sires used prior to 1994 and 58 calves born in 1992 and 1993.

USDA Line 1 Herefords. The Line 1 population of Hereford cattle was established at the Fort Keogh Livestock and Range Research Laboratory (Miles City, MT) as part of a series of projects in the western United States to produce inbred seedstock for exploiting heterosis in commercial production (MacNeil et al., 1992). The population was closed to outside introduction of germplasm in 1935, and selection for increased postweaning growth has continued since the inception of Line 1. Miles City Line 1 Herefords had a tremendous impact on the Hereford breed during the 1970s and 1980s when selection goals emphasized increased growth traits (Kuykendall, 1989). Advance Domino 20th and Advance Domino 54th, both sons of Advance Domino 13th, were used extensively in the formation of Miles City Line 1 (MacNeil et al., 1992). Relationships of Advance Domino 20th to the founding Lents Anxiety 4th sires Lord Lamplighter 8th and 3 Imperial Lamplighter 49th are .33 and .30, respectively, and relationships of Advance Domino 54th to the same Lents Anxiety 4th sires are .30 and .26. Although mating of close relatives was avoided, calves born in the Miles City Line 1 herd were 17% inbred by 1941, and the current average inbreeding is .30. The effective population size of Line 1 is 39.4, calculated as one-half of the rate of inbreeding (Falconer, 1989). Detailed trends of inbreeding as well as husbandry and management practices of Line 1 are described in MacNeil et al. (1992).

A total of 430 calves born in 1991, 1992, and 1993 was involved in this study, including 12 sires of 1991 and 1992 calves used to represent the population for initial genotyping. Direct EPD for birth weight (**BW-d**), 180-d gain from birth to weaning (**PR-d**), gain from weaning to yearling (**PO-d**), and maternal EPD for birth weight (**BW-m**) and 180-d gain from birth to weaning (**PR-m**) were estimated. Animal model analyses of birth weight, preweaning gain, and postweaning gain were conducted to predict EPD. Preweaning gain was the linear approximation of daily gain from birth to weaning multiplied by 180. Postweaning gain was the average daily gain observed during a postweaning gain test multiplied by 185. Separate analyses were performed for each trait. Pedigree records predate the establishment of Miles City Line 1 in 1934 by five generations. Performance records were edited to eliminate observations of twins and observations in contemporary groups of individual animals. Fixed effects were year-sex-age of dam subclasses and the linear regression on inbreeding of calf. Linear regressions on inbreeding of dam were also included in the analyses of birth weight and preweaning gain. All models included random effects of aggregate genotype of calf (direct effect) and residual. Analyses of birth weight and preweaning gain also included random effects of aggregate genotype of dam (maternal effect), the covariance of direct and maternal effects, and a permanent environmental effect due to dams. The analyses were conducted using REML procedures (Meyer, 1989) as programmed in MTDFREML (Boldman et al., 1993).

High and Low EPD Hereford Sires. Thirteen sires with high yearling weight (**YWT**) EPD and 14 sires with low YWT EPD were selected from the 1992 U.S. Hereford Sire Summary. The average YWT EPD of the high and low groups was +43.9 and +7.8, respectively, compared to the 1992 breed average of +21.8 (Benyshek et al., 1993). Accuracies for all YWT EPD were greater than .60. These sires were essentially non-inbred, with an average coefficient of .009 calculated from six generation pedigrees. The EPD sires were used in this study to represent commercial Hereford cattle throughout the United States with either high or low genetic potentials for growth.

DNA Extraction and Genotyping

Genomic DNA was purified from all animals from blood or semen samples using proteinase K digestion followed by either phenol-chloroform or high salt

extraction of proteins, and ethanol or isopropanol precipitation. Genotypes were determined for five PCR-based bi-allelic restriction fragment length polymorphisms within structural genes, including kappa-casein (**K-Cas**; Medrano and Aguilar-Cordova, 1990a), beta-lactoglobulin (**B-Lac**; Medrano and Aguilar-Cordova, 1990b), growth hormone (**GH**; Zhang et al., 1992), pituitary transcription factor 1 (**PIT1**; Moody et al., 1995a), and growth hormone receptor (**GHR**; Moody et al., 1995b). Microsatellite length polymorphisms were genotyped in the insulin-like growth factor I (**IGF-I**) and prolactin (**PRL**) loci. The IGF-I genotypes were determined using unique primers flanking the microsatellite described by Kirkpatrick (1992), with the 130- and 128-bp alleles designated as A and B, respectively. The PRL genotypes were determined using PCR primers designed to flank a microsatellite in the 5′ region of the bovine prolactin gene sequenced by Wolf et al. (1990). Two alleles, designated A and B, were observed. Genotypes for the bovine microsatellite BM2113 (Sunden et al., 1993) were determined in order to study a random, multi-allelic polymorphism not associated with a known structural gene.

The loci investigated are all located on different bovine chromosomes (Barendse et al., 1994; Moody et al., 1995a,b). Polymorphisms within the GH, K-Cas, and B-Lac genes are in coding regions and represent different forms of the proteins coded by those genes. Thus, the K-Cas, B-Lac, and GH polymorphisms have the potential of identifying a direct physiological effect resulting from differences in the amino acid sequences of their resulting proteins, as well as being markers of linked QTL. In contrast, polymorphisms in the PIT1, IGF-I, GHR, and PRL genes, as well as the BM2113 microsatellite, are located in non-coding regions and should only be considered as potential markers of linked QTL.

Statistical Analysis

Allele Frequencies and Variances. Allele frequencies were determined for each polymorphism in each population. Within the Lents population, only the eight herd sires were genotyped for the PRL, PIT1, IGF-I, and BM2113 polymorphisms because of little variation at these loci, and only the 12 herd sires representing the Line 1 population were genotyped for the PRL and BM2113 polymorphisms. Variances of allele frequencies were calculated according to Weir (1990).

Allele frequencies were compared using contingency table chi-square tests. Preliminary analyses compared allele frequencies between high and low YWT EPD sires. No significant differences were observed between the two groups for any loci, so they were pooled as one population (EPD sires) representing commercial Hereford sires with a wide range in genetic potential for growth traits. Further comparisons were made to evaluate differences in allele frequencies among Line 1, Lents Anxiety 4th, and EPD sire populations at each locus. A continuity correction factor of .5 was subtracted from the expected values in the numerator of the test statistic to account for using a continuous chi-square distribution to test hypotheses with discrete genotypic counts. Test statistics comparing three populations were compared for a .05 significance level with two degrees of freedom. If allele frequencies were significantly different among the three populations, then comparisons were made between pairs of populations. One degree of freedom was used to determine differences between two populations at the .05 significance level. Because the three pairwise tests are not completely independent, the overall error rate of these tests exceeds .05.

A heterozygosity coefficient ($\mathbf{H_l}$) was determined for each polymorphism in each population as the ratio of the number of heterozygotes to the total number of animals genotyped in that population. The H_l calculated for all polymorphisms were averaged within populations to estimate average genetic variation. The variance of H_l was calculated according to Weir (1990).

QTL Analysis in Miles City Line 1 Population. Regression analyses were performed in which BW-d, BW-m, PR-d, PR-m, and PO-d were the dependent variables, and genotype was the independent variable. Average effect of allele substitution was determined (α; Falconer, 1989) by coding genotypes as 0 (AA), 1 (AB), or 2 (BB) to represent the number of B alleles present for the K-Cas, B-Lac, GH, IGF-I, and PIT1 polymorphisms.

As described by Falconer (1989), the regression coefficient (α) estimates the average effect of allele substitution, or the average effect of replacing an A allele with a B allele. The y-intercept (a) estimates the average value of the AA genotype, and dominance deviations (d) are estimated by the difference between observed and predicted values at the AB genotype. The coefficient of determination (R^2) estimates the percentage of variability of the dependent variable that is explained by genotype.

Results

Allele and Genotype Frequencies. The K-Cas, B-Lac, GH, PIT1, IGF-I, and PRL A allele frequencies are shown in Figure 1, and BM2113 allele frequencies are shown in Figure 2. In the EPD sire population, both A and B alleles of the B-Lac, K-Cas, GH, PRL, PIT1, and IGF-I polymorphisms were segregating, and 5 of 11 known alleles of the BM2113 microsatellite marker were observed. In the Line 1 population, A and B alleles of the B-Lac, K-Cas, GH, PIT1, and IGF-I polymorphisms were segregating. The PRL polymorphism was fixed for the A allele, and only two alleles

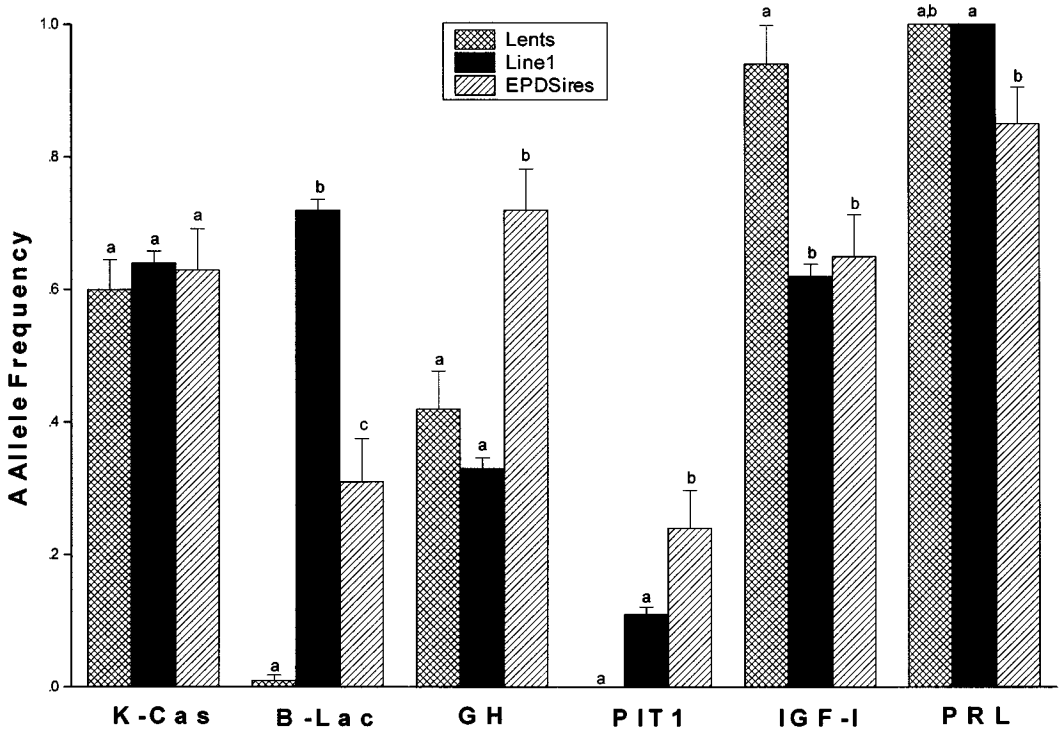

Figure 1. Allele frequencies of K-Cas, B-Lac, GH, PIT1, IGF-I, and PRL polymorphisms for three populations of Hereford cattle. At each locus, populations with different superscripts have different allele frequencies ($P < .05$).

(139 and 135 bp) of the BM2113 marker were present. In the Lents Anxiety 4th population, A and B alleles of the K-Cas and GH polymorphisms were segregating, but the PIT1 and PRL polymorphisms were fixed for the B and A alleles, respectively. The Lents population also seemed to be close to fixation for the B, A, and 139-bp alleles of the B-Lac, IGF-I, and BM2113 polymorphisms, respectively. The A allele of the GHR polymorphism was fixed in all populations, which is consistent with the observed lack of the B allele in *Bos taurus* cattle (Moody et al., 1995b).

Comparison of Allele Frequencies. Table 1 shows the contingency table chi-square test statistics that were calculated when testing for differences in allele frequencies among Line 1, Lents Anxiety 4th, and EPD sire populations. Results indicate that significant differences in allele frequencies among the three populations exist at all loci except K-Cas.

All three populations had significantly different allele frequencies for the B-Lac polymorphism. Allele frequencies of the EPD sire population were different from those of the Lents population at all loci except PRL and K-Cas and from those of the Line 1 population at all loci except IGF-I and K-Cas. Lents and Line 1 population allele frequencies differed only at the IGF-I and B-Lac loci.

Genetic Variation. The H_l and variances for each polymorphism in each population are shown in Figure 3, along with the average H_l for each population. The EPD sire population had the highest and the Lents

Table 1. Contingency table chi-square test statistics calculated for comparisons of allele frequencies among populations

Polymorphism	EPD/Line 1/Lents	Line 1/Lents	Line 1/EPD	Lents/EPD
B-Lac	238.63*	221.09*	38.87*	35.63*
K-Cas	.60	—	—	—
GH	34.55*	3.37	33.07*	13.35*
PRL	(8.29*)[a]	(.26)	(4.90*)	(3.49)
IGF	7.48*	7.00*	.56	(4.84*)
PIT1	10.07*	(3.08)	6.76*	(5.41*)
BM2113	(47.47*)	(4.40)	(24.02*)	(14.61*)

[a]Values shown in parentheses indicate at least one expected value in the analysis was less than five.
*$P < .05$.

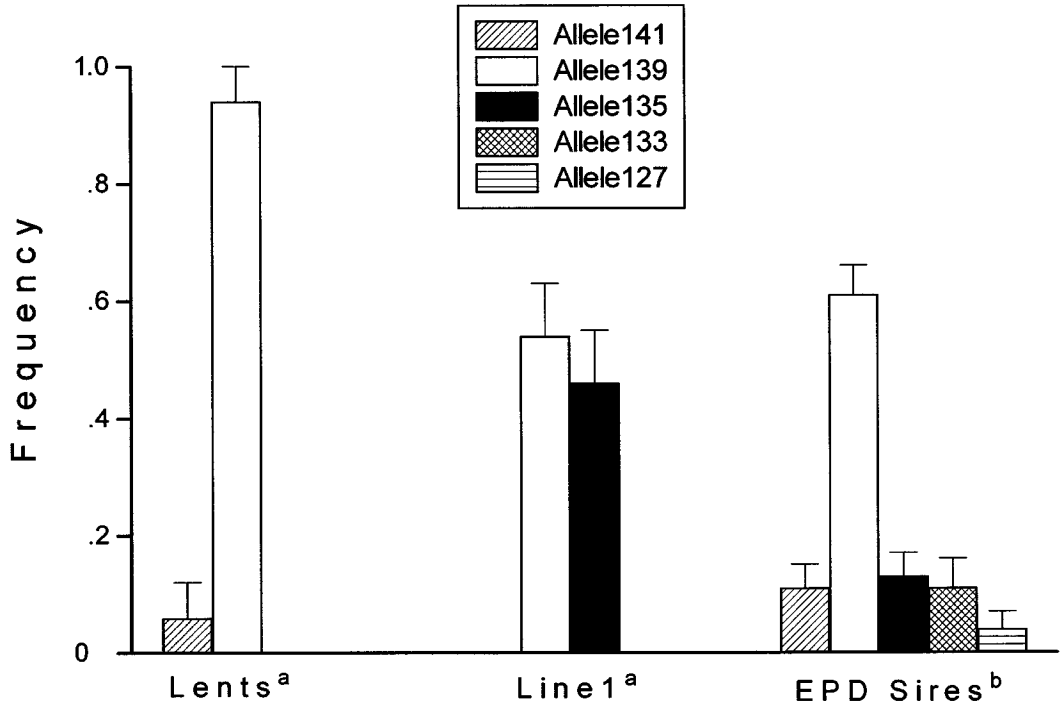

Figure 2. Allele frequencies of the BM2113 polymorphism for three populations of Hereford cattle. Populations with different superscripts have different allele frequencies ($P < .05$).

population the lowest average heterozygosity, as would be expected from the inbreeding coefficients. This trend was observed specifically at the IGF-I, PIT1, and BM2113 polymorphisms. Little variation was present at the B-Lac polymorphism in the Lents population ($H_{B\text{-}Lac} = .02$), but genetic variability was similar in the Line 1 ($H_{B\text{-}Lac} = .40$) and EPD sire ($H_{B\text{-}Lac} = .41$) populations at this locus. For the K-Cas and GH polymorphisms, the least amount of genetic variation was observed in the Line 1 population ($H_{K\text{-}Cas} = .38$; $H_{GH} = .46$). The EPD sire population had the greatest genetic variability at the K-Cas polymorphism ($H_{K\text{-}Cas} = .52$), whereas the Lents population had the greatest genetic variability at the GH polymorphism ($H_{GH} = .60$).

Regression Analysis. The means and standard deviations for EPD used in statistical analyses for markers of QTL in the Line 1 population are presented in Table 2. Results from the regression of BW-d, BW-m, PR-d, PR-m, and PO-d EPD on genotype are shown in Table 3. Substitution of a B allele for an A allele had significant effects on BW-d for the K-Cas, B-Lac, IGF-I, and PIT1 polymorphisms. Allele substitution of those polymorphisms, as well as GH, also had significant effects on BW-m. The effect of substituting a B allele for an A allele on PR-d was significant for K-Cas, B-Lac, and IGF-I polymorphisms. Allele substitution of K-Cas and IGF-I, as well as GH, also had significant effects on PR-m. The effect of allele substitution on PO-d was significant for the GH and IGF-I polymorphisms.

The percentage of variability in EPD explained by genotype when genotype effects were significant (R^2; Table 3) ranged from 1 to 16%. The greatest amounts of variability explained were for BW-d (IGF-I, 16%; K-Cas 15%) and PR-m (K-Cas, 8%). No more than 5% of variability was explained by any other polymorphism, even though several effects seemed statistically significant. Estimates of dominance deviations were significant only for the effect of IGF-I on BW-d and BW-m.

Discussion

Part I: Population Genetic Parameters

Allele frequencies differed among Hereford populations in this study at six of the seven polymorphisms

Table 2. Summary of Miles City Line 1 EPD included in QTL analyses

Trait	Mean, kg	Standard deviation
BW-d	.64	.85
BW-m	.32	.32
PR-d	5.67	1.47
PR-m	5.18	2.11
PO-d	6.37	3.30

genotyped. The observed differences likely resulted from a combination of factors including selection pressures, frequency differences in founder animals, and random genetic drift.

The Line 1 population has undergone continuous selection pressure for increased postweaning growth (MacNeil et al., 1992) but the Lents population has avoided such selection. Because of this, it was anticipated that loci influenced by selection for growth may have different allele frequencies between the populations. Allele frequencies in the Line 1 and Lents Anxiety 4th populations differed at the IGF-I and B-Lac polymorphisms. Differences in selection pressures for growth may have contributed to these differences in allele frequency, but this study cannot separate the effects of selection from confounded effects of other factors.

Differences in allele frequencies of founding animals of the Line 1 and Lents Anxiety 4th populations may also have contributed to present differences in gene frequencies. Relationships among the founding sires of the Lents Anxiety 4th (Lord Lamplighter 8th and 3 Imperial Lamplighter 49th) and Line 1 (Advance Domino 20th and Advance Domino 54th) populations range from .26 to .33, indicating these founding sires were related slightly more than half-sibs. Thus, the gene pools from which the two populations developed were likely similar. This hypothesis is supported by the observation of similar gene frequencies at five of the seven polymorphisms genotyped.

The observed differences in allele frequencies among the Line 1, Lents Anxiety 4th, and EPD sire populations most likely demonstrate the effects of random genetic drift acting in different directions or at different rates in the populations. Because the effects of drift are magnified in small populations, drift likely had a greater influence in the Lents Anxiety 4th population than in the Line 1 population. In contrast, the EPD sires were studied as a representation of the entire Hereford breed. Because of the broad genetic base they represent, allele frequencies in this group were likely less affected by drift.

The accumulation of inbreeding in the small Lents Anxiety 4th and Line 1 herds, combined with genetic drift, could lead to the loss of genetic variation. The random microsatellite marker BM2113 clearly shows the potential consequences of inbreeding, small population size, and random drift. Five alleles were present at this locus in the EPD sires, two alleles of approximately equal frequency were observed in Line 1, and the Lents Anxiety 4th population approached fixation of a single allele. The Lents Anxiety 4th population also approached fixation at four other loci genotyped. The fixation of alleles observed in the Lents Anxiety 4th population contrasts with the EPD sires, which were variable at all seven loci genotyped. This observation is consistent with lower inbreeding levels and the larger population represented by the EPD sires.

K-Cas A allele frequencies of Hereford populations in this study are lower than the frequency reported in Holstein (Medrano, 1990), but higher than the frequency reported in Jersey (Medrano and Aguilar-Cordova, 1990a). The B-Lac A allele frequency observed in EPD sires was similar to that reported in Holstein (Medrano, 1990), but extreme differences at this locus were observed among the three Hereford populations in this study. Zhang et al. (1993) reported similar allele frequencies among beef breeds at the GH locus. Growth hormone allele frequencies of the EPD sire population were similar to those previously reported, but the A allele frequency in the Line 1 and Lents populations was considerably lower. The observed IGF-I A allele frequencies in this study are much higher than the frequency reported by Kirkpatrick (1992) in a mixed-breed population. This study clearly demonstrates that significant within-breed as well as between-breed variation in allele frequencies exists among cattle populations.

Differences in allele frequencies indicate the possibility of differences in genetic backgrounds among

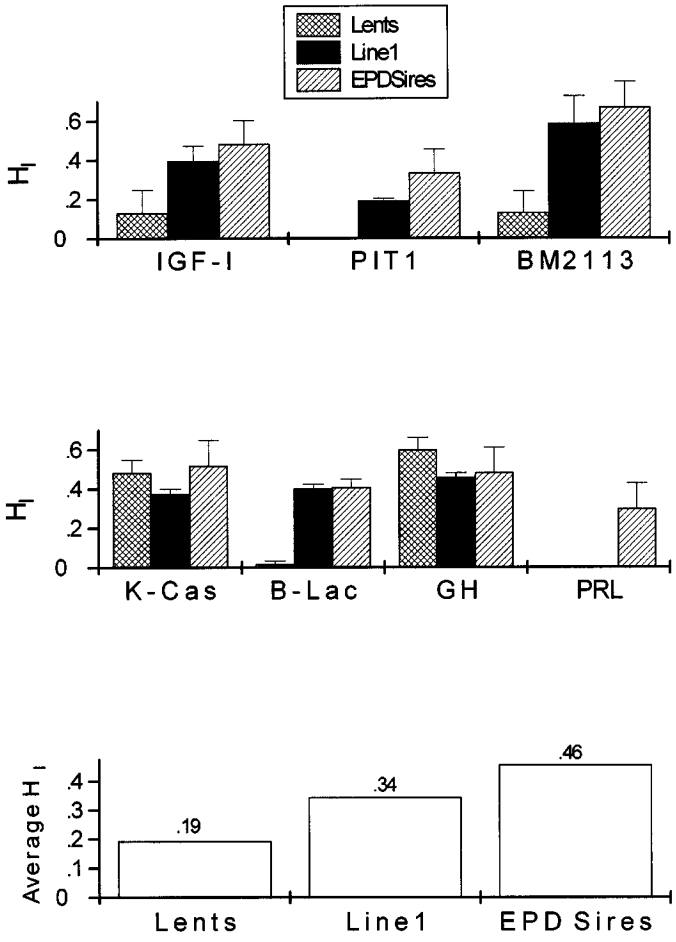

Figure 3. Heterozygosity coefficients (H_I) for three populations of Hereford cattle.

Table 3. Results from regression analyses of direct EPD for birth weight (BW-d), maternal EPD for birth weight (BW-m), direct EPD for 180-d gain from birth to weaning (PR-d), maternal EPD for 180-d gain from birth to weaning (PR-m), and direct EPD for gain from weaning to yearling (PO-d) on genotype

	a[a]	α[b]	P (α = 0)	d[c]	P (d = 0)	R^2, %[d]
K-Cas (n = 409)						
BW-d	.958 ± .054	–.421 ± .055	<.01	–.098 ± .084	.24	15
BW-m	.381 ± .020	–.075 ± .020	<.01	–.022 ± .031	.47	4
PR-d	5.881 ± .100	–.298 ± .102	<.01	–.211 ± .156	.18	3
PR-m	5.338 ± .140	–.820 ± .124	<.01	–.052 ± .217	.81	8
PO-d	6.634 ± .229	–.376 ± .233	.10	.018 ± .356	.96	1
B-Lac (n = 403)						
BW-d	.765 ± .055	–.228 ± .065	<.01	–.066 ± .102	.52	3
BW-m	.353 ± .019	–.051 ± .023	.05	.019 ± .036	.60	1
PR-d	5.456 ± .097	.318 ± .114	<.01	.004 ± .182	.98	2
PR-m	5.170 ± .140	.128 ± .166	.44	.461 ± .261	.08	
PO-d	6.302 ± .219	.042 ± .260	.87	.101 ± .410	.80	
GH (n = 408)						
BW-d	.567 ± .095	.062 ± .064	.34	.088 ± .095	.36	
BW-m	.403 ± .033	–.057 ± .022	<.01	.015 ± .033	.64	2
PR-d	5.861 ± .164	–.166 ± .110	.13	.153 ± .165	.36	
PR-m	4.303 ± .232	.690 ± .156	<.01	.264 ± .233	.26	5
PO-d	5.121 ± .370	.887 ± .248	<.01	.405 ± .371	.28	3
IGF-I (n = 406)						
BW-d	.314 ± .055	.444 ± .053	<.01	.167 ± .083	.04	16
BW-m	.279 ± .021	.065 ± .020	<.01	.066 ± .031	.03	4
PR-d	5.308 ± .101	.431 ± .097	<.01	–.273 ± .152	.07	5
PR-m	4.914 ± .148	.423 ± .141	<.01	–.284 ± .222	.20	3
PO-d	6.697 ± .236	–.452 ± .224	.04	–.413 ± .353	.24	1
PIT1 (n = 410)						
BW-d	1.272 ± .162	–.353 ± .088	<.01	.233 ± .176	.19	4
BW-m	.507 ± .056	–.102 ± .031	<.01	–.016 ± .061	.80	3
PR-d	5.740 ± .287	–.062 ± .157	.69	.091 ± .312	.77	
PR-m	5.341 ± .413	.070 ± .226	.76	.361 ± .449	.42	
PO-d	7.489 ± .650	–.651 ± .355	.07	.503 ± .708	.48	1

[a]Average EPD of the AA genotype.
[b]Average effect of allele substitution (kg).
[c]Dominance deviation (kg).
[d]Percentage of variability of EPD explained by genotype.

populations. The genetic background of a population may prove to be an important factor as DNA markers linked to QTL are identified. Quantitative trait loci influencing a trait in one population may have a different effect, or no effect at all, in another population due to epistatic interactions of the QTL with background genes (Pomp, 1994).

Heterozygosity. It was expected that the Lents population would have the lowest H_l and that the pooled EPD population would have the highest H_l because of known inbreeding levels. These expectations were verified at the IGF-I, PRL, BM2113, and PIT1 polymorphisms, but not at the K-Cas, B-Lac, or GH polymorphisms. Selection or drift may be preventing the latter loci from displaying the expected consequences of inbreeding.

If the H_l of the EPD sire population is assumed to be similar to the H_l of the founding populations of the Lents Anxiety 4th and Line 1 herds, decreases of 54% and 30% in the Lents and Line 1 populations, respectively, would be expected based on known inbreeding levels. The average H_l of the Lents and Line 1 populations actually decreased by 59% and 26%, respectively, relative to the average H_l of the EPD sire population. Thus, the average H_l closely reflects the decrease in heterozygosity that is expected due to inbreeding, even though some individual loci failed to meet expectations.

Expectations of Hardy-Weinberg Equilibrium (**HWE**) were tested in the Line 1 and Lents Anxiety 4th populations at five and two loci, respectively (analyses not shown). Surprisingly, expectations were met at three loci in Line 1, and at both loci tested in the Lents Anxiety 4th population. These results were unexpected because several underlying assumptions of HWE were violated (Falconer, 1989).

Part II: Markers Associated with QTL

At the time this project was initiated, bovine genetic linkage maps (Barendse et al., 1994; Bishop et al., 1994) were not yet available. Consequently, a

candidate gene approach was utilized, and genes involved in the growth hormone axis (GH, GHR, PIT1, and IGF-I) and milk production (K-Cas, B-Lac, and PRL) were selected. The GHR and PRL polymorphisms were not informative markers in this study because the A allele of both loci was fixed in the Line 1 population.

Regression analysis identified several significant effects of genotype on EPD in the Line 1 population. The largest effects were those of IGF-I on BW-d and K-Cas on BW-d and PR-m, which explained 16, 15, and 8% of variability, respectively. Unfortunately, substitution of the B allele for an A allele of K-Cas resulted in the desirable effect of decreasing BW-d along with the often undesirable effect of decreasing PR-m.

In addition to regression of EPD on genotype, regression analyses using actual birth weight, weaning weight, and yearling weight phenotypic data adjusted for year, sex, and age of dam were completed. Results (data not shown) were very similar to those shown in Table 3. Phenotypic data were also analyzed using an animal model to account for relationships among animals. Contrasts testing for additive and dominant gene action failed to detect significant effects of genotype on birth weight, weaning weight, or yearling weight (data not shown).

Milk production accounts for a major portion of PR-m. Therefore, PR-m may be considered as an estimate of milk production of the Line 1 dams. Previous studies have considered the effects of milk protein genes, including K-Cas and B-Lac, on milk production traits of dairy cattle. In contrast to the present results, several studies have reported a trend for an increase in milk yield associated with the K-Cas B allele (Lin et al., 1989; Van Eenennaam and Medrano, 1991; Cowan et al., 1992). However, others have reported trends of higher milk yields from cows with the K-Cas A allele (Ng-Kwai-Hang et al., 1986; Gonyon et al., 1987; Bovenhuis et al., 1992; Bovenhuis and Weller, 1994). The K-Cas gene is closely linked to the β-casein (**B-Cas**) gene (Hines et al., 1981; Threadgill and Womack, 1990). Bovenhuis et al. (1992) and Bovenhuis and Weller (1994) showed that the observed effect of K-Cas on milk yield in the Dutch dairy cattle population actually resulted from linkage to the B-Cas gene. Previous studies had not distinguished between direct effects of milk protein genes and effects of linked QTL. Therefore, conflicting reports of the effects of K-Cas genotype on milk yield may be explained by differences in linkage disequilibrium between K-Cas and B-Cas in different populations. Additionally, differences in genetic background may also influence the effects of this QTL in different populations.

Milk composition, as well as milk production, may contribute to PR-m. Several studies have found a significant increase in milk protein percentage associated with the K-Cas B allele in Holstein cattle (Ng-Kwai-Hang et al., 1986; Gonyon et al., 1987; Aleandri et al., 1990; Bovenhuis et al., 1992; Bovenhuis and Weller, 1994). Therefore, the observed differences in PR-m among K-Cas genotypes in this study may be due to differences in milk composition as well as milk yield.

A significant effect of B-Lac on milk yield has been reported, with increases consistently associated with the A allele (Geldermann et al., 1985; Aleandri et al., 1990; Bovenhuis et al., 1992; Cowan et al., 1992; Bovenhuis and Weller, 1994). Bovenhuis et al. (1992) and Bovenhuis and Weller (1994) also reported a significant increase in fat percentage associated with the B allele. A QTL influencing protein percentage and yield closely linked to B-Lac was also identified (Bovenhuis et al., 1992; Bovenhuis and Weller, 1994). Results from the present study failed to detect a significant effect of the B-Lac polymorphism on PR-m.

The GH polymorphism investigated in this study results from a single point mutation that causes either a leucine (A allele) or valine (B allele) to be incorporated at amino acid position 127 of GH (Seavey et al., 1971; Zhang et al., 1992). Eppard et al. (1992) reported higher milk yields from dairy cows that received recombinant-derived GH with valine at position 127, compared to cows receiving GH with leucine at position 127. These results are consistent with the present study, in which the GH B allele was associated with a significant increase in PR-m, explaining 5% of PR-m variability.

The structure of the Line 1 population, which included several small half-sib families, is far from ideal for a study designed to identify QTL, especially when using non-coding region markers such as IGF-I and PIT1. More efficient and appropriate experimental designs have been proposed (Weller et al., 1990; Keele, 1994) but were not possible with existing resources. As a result, the power of the statistical analysis in this study was limited and significant effects explaining small amounts of variation are likely spurious. The large effect of IGF-I on BW-d should also be interpreted with caution because this study was unable to follow the segregation of IGF-I alleles in large half-sib families. However, the effects of K-Cas genotype on BW-d and PR-m may indicate a direct effect of the K-Cas gene or of a very closely linked gene. The magnitude of these effects, as well as evidence that the K-Cas polymorphism is linked to a QTL influencing milk production in dairy cattle (Bovenhuis et al., 1992; Cowan et al., 1992; Bovenhuis and Weller, 1994), indicates K-Cas genotype may be a useful marker for BW-d and(or) PR-m in future marker-assisted selection programs in Hereford cattle.

Implications

Significant differences in allele frequencies were found among three populations of Hereford cattle at six of seven DNA polymorphisms evaluated, indicating

potential differences in genetic backgrounds exist among populations within breeds. In Miles City Line 1 Herefords, a polymorphism in the kappa-casein gene accounted for 15 and 8% of the variability in expected progeny differences for birth weight and 180-d gain from birth to weaning, respectively, indicating that kappa-casein may be a useful marker for these traits in future marker-assisted selection programs in Hereford cattle.

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